```
  * * * * * * * * * * * * * STN  Columbus  * * * * * * * * * * * * * * *

FILE 'HOME' ENTERED AT 15:29:07 ON 04 APR 2005

=> fil .bec
COST IN U.S. DOLLARS                                  SINCE FILE      TOTAL
                                                         ENTRY    SESSION
FULL ESTIMATED COST                                       0.21       0.21

FILES 'MEDLINE, SCISEARCH, LIFESCI, BIOTECHDS, BIOSIS, EMBASE, HCAPLUS, NTIS,
       ESBIOBASE, BIOTECHNO, WPIDS' ENTERED AT 15:29:50 ON 04 APR 2005
ALL COPYRIGHTS AND RESTRICTIONS APPLY.  SEE HELP USAGETERMS FOR DETAILS.

11 FILES IN THE FILE LIST

=> s vitro(8a)sialyl?
FILE 'MEDLINE'
        791974 VITRO
          6892 SIALYL?
L1          95 VITRO(8A)SIALYL?

FILE 'SCISEARCH'
        419411 VITRO
          7180 SIALYL?
L2          78 VITRO(8A)SIALYL?

FILE 'LIFESCI'
        186324 VITRO
          1690 SIALYL?
L3          26 VITRO(8A)SIALYL?

FILE 'BIOTECHDS'
         25126 VITRO
           453 SIALYL?
L4           5 VITRO(8A)SIALYL?

FILE 'BIOSIS'
        630792 VITRO
          7595 SIALYL?
L5         106 VITRO(8A)SIALYL?

FILE 'EMBASE'
        937897 VITRO
          6552 SIALYL?
L6          89 VITRO(8A)SIALYL?

FILE 'HCAPLUS'
        590032 VITRO
          8846 SIALYL?
L7         128 VITRO(8A)SIALYL?

FILE 'NTIS'
          8795 VITRO
            18 SIALYL?
L8           0 VITRO(8A)SIALYL?

FILE 'ESBIOBASE'
        188679 VITRO
          2946 SIALYL?
L9          49 VITRO(8A)SIALYL?

FILE 'BIOTECHNO'
        253158 VITRO
          3202 SIALYL?
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L10          58 VITRO(8A)SIALYL?

FILE 'WPIDS'
         22785 VITRO
           471 SIALYL?
L11            5 VITRO(8A)SIALYL?

TOTAL FOR ALL FILES
L12          639 VITRO(8A) SIALYL?

=> s l12 not 1998-2005/py
FILE 'MEDLINE'
        3745483 1998-2005/PY
L13           65 L1 NOT 1998-2005/PY

FILE 'SCISEARCH'
        7277942 1998-2005/PY
L14           41 L2 NOT 1998-2005/PY

FILE 'LIFESCI'
         749707 1998-2005/PY
L15           16 L3 NOT 1998-2005/PY

FILE 'BIOTECHDS'
         138819 1998-2005/PY
L16            4 L4 NOT 1998-2005/PY

FILE 'BIOSIS'
        3819440 1998-2005/PY
L17           70 L5 NOT 1998-2005/PY

FILE 'EMBASE'
        3313559 1998-2005/PY
L18           61 L6 NOT 1998-2005/PY

FILE 'HCAPLUS'
        6920095 1998-2005/PY
L19           79 L7 NOT 1998-2005/PY

FILE 'NTIS'
         138251 1998-2005/PY
L20            0 L8 NOT 1998-2005/PY

FILE 'ESBIOBASE'
        2083977 1998-2005/PY
L21           19 L9 NOT 1998-2005/PY

FILE 'BIOTECHNO'
         724097 1998-2005/PY
L22           36 L10 NOT 1998-2005/PY

FILE 'WPIDS'
        5843979 1998-2005/PY
L23            0 L11 NOT 1998-2005/PY

TOTAL FOR ALL FILES
L24          391 L12 NOT 1998-2005/PY

=> s (commercial or scale or batch)(10a)(sialyl? or glycosylat?)
FILE 'MEDLINE'
          44012 COMMERCIAL
         140218 SCALE
          11056 BATCH
           6892 SIALYL?
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         43583 GLYCOSYLAT?
L25         63 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'SCISEARCH'
         99675 COMMERCIAL
        321971 SCALE
         38411 BATCH
          7180 SIALYL?
         34005 GLYCOSYLAT?
L26         102 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'LIFESCI'
         23050 COMMERCIAL
         35704 SCALE
         11006 BATCH
          1690 SIALYL?
         10133 GLYCOSYLAT?
L27         28 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'BIOTECHDS'
          6189 COMMERCIAL
         15425 SCALE
         12791 BATCH
           453 SIALYL?
          4005 GLYCOSYLAT?
L28         68 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'BIOSIS'
         95417 COMMERCIAL
        153467 SCALE
         24881 BATCH
          7595 SIALYL?
         36797 GLYCOSYLAT?
L29         78 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'EMBASE'
         41283 COMMERCIAL
        153218 SCALE
         16987 BATCH
          6552 SIALYL?
         34480 GLYCOSYLAT?
L30         60 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'HCAPLUS'
         29853 COMMERCIAL
        283685 COM
        297846 COMMERCIAL
                 (COMMERCIAL OR COM)
        346358 SCALE
         85128 BATCH
          8846 SIALYL?
         41426 GLYCOSYLAT?
L31        167 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'NTIS'
         53181 COMMERCIAL
         82474 SCALE
          6385 BATCH
            18 SIALYL?
           124 GLYCOSYLAT?
L32          1 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'ESBIOBASE'
         24125 COMMERCIAL
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         60287 SCALE
         11804 BATCH
          2946 SIALYL?
         13486 GLYCOSYLAT?
L33         50 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'BIOTECHNO'
         14938 COMMERCIAL
         23003 SCALE
         11409 BATCH
          3202 SIALYL?
         16990 GLYCOSYLAT?
L34         46 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

FILE 'WPIDS'
         45427 COMMERCIAL
        129581 SCALE
         28080 BATCH
           471 SIALYL?
          2880 GLYCOSYLAT?
L35         13 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

TOTAL FOR ALL FILES
L36        676 (COMMERCIAL OR SCALE OR BATCH)(10A)(SIALYL? OR GLYCOSYLAT?)

=> s l36 not 1998-2005/py
FILE 'MEDLINE'
       3745483 1998-2005/PY
L37         30 L25 NOT 1998-2005/PY

FILE 'SCISEARCH'
       7277942 1998-2005/PY
L38         45 L26 NOT 1998-2005/PY

FILE 'LIFESCI'
        749707 1998-2005/PY
L39         15 L27 NOT 1998-2005/PY

FILE 'BIOTECHDS'
        138819 1998-2005/PY
L40         47 L28 NOT 1998-2005/PY

FILE 'BIOSIS'
       3819440 1998-2005/PY
L41         42 L29 NOT 1998-2005/PY

FILE 'EMBASE'
       3313559 1998-2005/PY
L42         31 L30 NOT 1998-2005/PY

FILE 'HCAPLUS'
       6920095 1998-2005/PY
L43         74 L31 NOT 1998-2005/PY

FILE 'NTIS'
        138251 1998-2005/PY
L44          1 L32 NOT 1998-2005/PY

FILE 'ESBIOBASE'
       2083977 1998-2005/PY
L45         12 L33 NOT 1998-2005/PY

FILE 'BIOTECHNO'
        724097 1998-2005/PY
```

L46 24 L34 NOT 1998-2005/PY

FILE 'WPIDS'
5843979 1998-2005/PY
L47 3 L35 NOT 1998-2005/PY

TOTAL FOR ALL FILES
L48 324 L36 NOT 1998-2005/PY

=> d l37 1

L37 ANSWER 1 OF 30 MEDLINE on STN
TI Evaluation of a **commercial** kit for measurement of **glycosylated** hemoglobin in canine blood.
SO Veterinary clinical pathology / American Society for Veterinary Clinical Pathology, (1981) 10 (1) 21-4.
Journal code: 9880575. ISSN: 0275-6382.
AU Mahaffey E A; Cornelius L M
AN 2004406891 IN-PROCESS

=> d tot l37

L37 ANSWER 1 OF 30 MEDLINE on STN
TI Evaluation of a **commercial** kit for measurement of **glycosylated** hemoglobin in canine blood.
SO Veterinary clinical pathology / American Society for Veterinary Clinical Pathology, (1981) 10 (1) 21-4.
Journal code: 9880575. ISSN: 0275-6382.
AU Mahaffey E A; Cornelius L M
AN 2004406891 IN-PROCESS

L37 ANSWER 2 OF 30 MEDLINE on STN
TI Chemoenzymatic synthesis of a trimeric ganglioside GM3 analogue.
SO Carbohydrate research, (1997 Jun 11) 301 (1-2) 1-4.
Journal code: 0043535. ISSN: 0008-6215.
AU Earle M A; Manku S; Hultin P G; Li H; Palcic M M
AN 97372515 MEDLINE

L37 ANSWER 3 OF 30 MEDLINE on STN
TI Exploring the substrate specificities of alpha-2,6- and alpha-2,3-sialyltransferases using synthetic acceptor analogues.
SO European journal of biochemistry / FEBS, (1996 Dec 15) 242 (3) 674-81.
Journal code: 0107600. ISSN: 0014-2956.
AU Van Dorst J A; Tikkanen J M; Krezdorn C H; Streiff M B; Berger E G; Van Kuik J A; Kamerling J P; Vliegenthart J F
AN 97175036 MEDLINE

L37 ANSWER 4 OF 30 MEDLINE on STN
TI Large-**scale** expression of recombinant **sialyltransferases** and comparison of their kinetic properties with native enzymes.
SO Glycoconjugate journal, (1995 Dec) 12 (6) 755-61.
Journal code: 8603310. ISSN: 0282-0080.
AU Williams M A; Kitagawa H; Datta A K; Paulson J C; Jamieson J C
AN 96318012 MEDLINE

L37 ANSWER 5 OF 30 MEDLINE on STN
TI Enlarged **scale** chemical synthesis and range of activity of drosocin, an O-**glycosylated** antibacterial peptide of Drosophila.
SO European journal of biochemistry / FEBS, (1996 May 15) 238 (1) 64-9.
Journal code: 0107600. ISSN: 0014-2956.
AU Bulet P; Urge L; Ohresser S; Hetru C; Otvos L Jr
AN 96248422 MEDLINE

L37 ANSWER 6 OF 30 MEDLINE on STN
TI Oligosaccharide mapping reveals hormone-specific glycosylation patterns on equine gonadotropin alpha-subunit Asn56.
SO Endocrinology, (1996 Jun) 137 (6) 2543-57.
Journal code: 0375040. ISSN: 0013-7227.
AU Gotschall R R; Bousfield G R
AN 96217295 MEDLINE

L37 ANSWER 7 OF 30 MEDLINE on STN
TI Separation of human serum transferrin isoforms by high-performance pellicular anion-exchange chromatography.
SO Protein expression and purification, (1996 Feb) 7 (1) 39-44.
Journal code: 9101496. ISSN: 1046-5928.
AU Rohrer J S; Avdalovic N
AN 96209930 MEDLINE

L37 ANSWER 8 OF 30 MEDLINE on STN
TI Recombinant soluble Fc gamma receptors: production, purification and biological activities.
SO Journal of chromatography. B, Biomedical applications, (1994 Dec 9) 662 (2) 197-207.
Journal code: 9421796. ISSN: 0378-4347.
AU Sautes C; Galinha A; Bouchard C; Mazieres N; Spagnoli R; Fridman W H
AN 95235775 MEDLINE

L37 ANSWER 9 OF 30 MEDLINE on STN
TI The macroheterogeneity of recombinant human interferon-gamma produced by Chinese-hamster ovary cells is affected by the protein and lipid content of the culture medium.
SO Biotechnology and applied biochemistry, (1995 Feb) 21 (Pt 1) 87-100.
Journal code: 8609465. ISSN: 0885-4513.
AU Castro P M; Ison A P; Hayter P M; Bull A T
AN 95225997 MEDLINE

L37 ANSWER 10 OF 30 MEDLINE on STN
TI Glycosylation of recombinant human granulocyte colony stimulating factor: implications for stability and potency.
SO European journal of cancer (Oxford, England : 1990), (1994) 30A Suppl 3 S12-4.
Journal code: 9005373. ISSN: 0959-8049.
AU Nissen C
AN 95209924 MEDLINE

L37 ANSWER 11 OF 30 MEDLINE on STN
TI Effect of lipid supplements on the production and glycosylation of recombinant interferon-gamma expressed in CHO cells.
SO Cytotechnology, (1994) 15 (1-3) 209-15.
Journal code: 8807027. ISSN: 0920-9069.
AU Jenkins N; Castro P; Menon S; Ison A; Bull A
AN 95200739 MEDLINE

L37 ANSWER 12 OF 30 MEDLINE on STN
TI Expression of mouse Gal beta 1,4GlcNAc alpha 2,6-sialyltransferase in an insoluble form in Escherichia coli and partial renaturation.
SO Bioorganic & medicinal chemistry, (1994 Feb) 2 (2) 79-84.
Journal code: 9413298. ISSN: 0968-0896.
AU Hamamoto T; Lee Y C; Kurosawa N; Nakaoka T; Kojima N; Tsuji S
AN 95006421 MEDLINE

L37 ANSWER 13 OF 30 MEDLINE on STN
TI Expression of glycosylated and nonglycosylated human transferrin in mammalian cells. Characterization of the recombinant proteins with comparison to three commercially available transferrins.
SO Biochemistry, (1993 May 25) 32 (20) 5472-9.

Journal code: 0370623. ISSN: 0006-2960.
AU Mason A B; Miller M K; Funk W D; Banfield D K; Savage K J; Oliver R W; Green B N; MacGillivray R T; Woodworth R C
AN 93271170 MEDLINE

L37 ANSWER 14 OF 30 MEDLINE on STN
TI [Expression of recombinant proteins in the milk of transgenic animals]. Expression de proteines recombinantes dans le lait d'animaux transgeniques.
SO Revue francaise de transfusion et d'hemobiologie : bulletin de la Societe nationale de transfusion sanguine, (1993 Jan) 36 (1) 49-72. Ref: 85
Journal code: 8908966. ISSN: 1140-4639.
AU Houdebine L M
AN 93236665 MEDLINE

L37 ANSWER 15 OF 30 MEDLINE on STN
TI Measurement of non-enzymic glycosylation with a radiochemical assay.
SO International journal of biochemistry, (1993 Mar) 25 (3) 379-84.
Journal code: 0250365. ISSN: 0020-711X.
AU Sheikh M A; Robb D A
AN 93215902 MEDLINE

L37 ANSWER 16 OF 30 MEDLINE on STN
TI Psychosocial and psychopathologic influences on management and control of insulin-dependent diabetes.
SO International journal of psychiatry in medicine, (1992) 22 (2) 105-17.
Journal code: 0365646. ISSN: 0091-2174.
AU Eaton W W; Mengel M; Mengel L; Larson D; Campbell R; Montague R B
AN 92387853 MEDLINE

L37 ANSWER 17 OF 30 MEDLINE on STN
TI The use of porcine liver (2----3)-alpha-**sialyltransferase** in the large-**scale** synthesis of alpha-Neup5Ac-(2----3)-beta-D-Galp-(1---3)-D-GlcpNAc, the epitope of the tumor-associated carbohydrate antigen CA 50.
SO Carbohydrate research, (1992 Apr 10) 228 (1) 137-44.
Journal code: 0043535. ISSN: 0008-6215.
AU Lubineau A; Auge C; Francois P
AN 92386536 MEDLINE

L37 ANSWER 18 OF 30 MEDLINE on STN
TI A modified quality-of-life measure for youths: psychometric properties.
SO Diabetes educator, (1991 Mar-Apr) 17 (2) 114-8.
Journal code: 7701401. ISSN: 0145-7217.
AU Ingersoll G M; Marrero D G
AN 91138475 MEDLINE

L37 ANSWER 19 OF 30 MEDLINE on STN
TI Recombinant human interferon-gamma. Differences in **glycosylation** and proteolytic processing lead to heterogeneity in **batch** culture.
SO Biochemical journal, (1990 Dec 1) 272 (2) 333-7.
Journal code: 2984726R. ISSN: 0264-6021.
AU Curling E M; Hayter P M; Baines A J; Bull A T; Gull K; Strange P G; Jenkins N
AN 91097442 MEDLINE

L37 ANSWER 20 OF 30 MEDLINE on STN
TI The use of immobilised glycosyltransferases in the synthesis of sialyloligosaccharides.
SO Carbohydrate research, (1990 Apr 25) 200 257-68.
Journal code: 0043535. ISSN: 0008-6215.
AU Auge C; Fernandez-Fernandez R; Gautheron C
AN 90335877 MEDLINE

L37 ANSWER 21 OF 30 MEDLINE on STN
TI Non-enzymatic glycosylation.
SO British medical bulletin, (1989 Jan) 45 (1) 174-90. Ref: 51
Journal code: 0376542. ISSN: 0007-1420.
AU Kennedy L; Lyons T J
AN 90002114 MEDLINE

L37 ANSWER 22 OF 30 MEDLINE on STN
TI Serological responses in cats vaccinated with FeLV ISCOM and an inactivated FeLV vaccine.
SO Vaccine, (1989 Apr) 7 (2) 137-41.
Journal code: 8406899. ISSN: 0264-410X.
AU Osterhaus A; Weijer K; UytdeHaag F; Knell P; Jarrett O; Akerblom L; Morein B
AN 89319150 MEDLINE

L37 ANSWER 23 OF 30 MEDLINE on STN
TI Constitutive long-term production and characterization of recombinant human interferon-gammas from two different mammalian cells.
SO Cell structure and function, (1988 Apr) 13 (2) 143-59.
Journal code: 7608465. ISSN: 0386-7196.
AU Sano E; Okano K; Sawada R; Naruto M; Sudo T; Kamata K; Iizuka M; Kobayashi S
AN 88253448 MEDLINE

L37 ANSWER 24 OF 30 MEDLINE on STN
TI Biotin binding to avidin. Oligosaccharide side chain not required for ligand association.
SO Biochemical journal, (1987 Nov 15) 248 (1) 167-71.
Journal code: 2984726R. ISSN: 0264-6021.
AU Hiller Y; Gershoni J M; Bayer E A; Wilchek M
AN 88133839 MEDLINE

L37 ANSWER 25 OF 30 MEDLINE on STN
TI Affinity chromatography: a precise method for glycosylated albumin estimation.
SO Annals of clinical biochemistry, (1985 Jan) 22 (Pt 1) 79-83.
Journal code: 0324055. ISSN: 0004-5632.
AU John W G; Jones A E
AN 85173127 MEDLINE

L37 ANSWER 26 OF 30 MEDLINE on STN
TI Comparison of five **commercial** kits for the determination of **glycosylated** haemoglobin.
SO Journal of clinical pathology, (1984 Oct) 37 (10) 1177-81.
Journal code: 0376601. ISSN: 0021-9746.
AU Norcliffe D; Turner E M
AN 85031129 MEDLINE

L37 ANSWER 27 OF 30 MEDLINE on STN
TI Comparison of four commercial methods for the determination of fast hemoglobins.
SO Clinical biochemistry, (1982 Aug) 15 (4) 230-3.
Journal code: 0133660. ISSN: 0009-9120.
AU Lee L P; Arnott B; Feng M; Hynie I
AN 83025558 MEDLINE

L37 ANSWER 28 OF 30 MEDLINE on STN
TI New chromatographic system for the rapid analysis and preparation of colostrum sialyloligosaccharides.
SO Journal of chromatography, (1981 Aug 7) 212 (3) 313-22.
Journal code: 0427043. ISSN: 0021-9673.
AU Veh R W; Michalski J C; Corfield A P; Sander-Wewer M; Gies D; Schauer R

AN 81264524 MEDLINE

L37 ANSWER 29 OF 30 MEDLINE on STN
TI A temperature conversion nomogram for glycosylated hemoglobin analysis.
SO Clinica chimica acta; international journal of clinical chemistry, (1980 Jun 10) 104 (2) 251-7.
Journal code: 1302422. ISSN: 0009-8981.
AU Hankins W D; Holladay L
AN 80223379 MEDLINE

L37 ANSWER 30 OF 30 MEDLINE on STN
TI A simple microchromatographic column for determination of hemoglobins A1a + b and A1c.
SO Hemoglobin, (1979) 3 (6) 411-28.
Journal code: 7705865. ISSN: 0363-0269.
AU Friedman S; Humbert J R
AN 80071587 MEDLINE

=> log y

COST IN U.S. DOLLARS	SINCE FILE ENTRY	TOTAL SESSION
FULL ESTIMATED COST	42.33	325.98

DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)	SINCE FILE ENTRY	TOTAL SESSION
CA SUBSCRIBER PRICE	-2.19	-2.19

STN INTERNATIONAL LOGOFF AT 16:11:07 ON 04 APR 2005

Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
L2	43	vitro near4 sialyla$	US-PGPUB; USPAT	OR	OFF	2005/04/04 14:38
L3	44	(commercial or scale or batch) near4 sialyl$	US-PGPUB; USPAT	OR	OFF	2005/04/04 15:00

Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
L2	43	vitro near4 sialyla$	US-PGPUB; USPAT	OR	OFF	2005/04/04 14:38

PGPUB-DOCUMENT-NUMBER: 20050031584

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20050031584 A1

TITLE: Interleukin-2:remodeling and glycoconjugation of interleukin-2

PUBLICATION-DATE: February 10, 2005

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
DeFrees, Shawn	North Wales	PA	US	
Zopf, David	Wayne	PA	US	
Bayer, Robert	San Diego	CA	US	
Bowe, Caryn	Doylestown	PA	US	
Hakes, David	Willow Grove	PA	US	
Chen, Xi	Lansdale	PA	US	

APPL-NO: 10/ 410980

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10410980 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10410980 A1 20030409

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10410980 A1 20030409

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

non-provisional-of-provisional 60344692 20011019 US

non-provisional-of-provisional 60328523 20011010 US

US-CL-CURRENT: 424/85.2, 530/351

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (1059):
[1381] In vitro sialylation was carried out using the G2 glycoform antibody (1 mg/mL) by contacting it with 0.1 U/mg ST3Gal3 or 0.1 U/mg ST6Gal1, 5 mM CMP-sialic acid, at 32.degree. C. for 24 hr in a buffer of 50 mM Tris pH 7.4, 150 mM NaCl, and 3 mM CMP-SA. An aliquot was removed for glycan analysis, and the remaining products were affinity purified as described above.

Detail Description Paragraph - DETX (1331):
[1653] Sialylation. TPA expressed in mammalian cells will often contain a majority of the glycans terminating in sialic acid, but to ensure complete sialylation, it would be beneficial to first perform an in vitro sialylation. TPA in a suitable buffer (most preferably between pH 5.5 and 9, for example Tris buffered saline, pH 7.2) is incubated with CMP sialic acid and sialyltransferase for a time sufficient to convert any glycans lacking sialic acid to sialylated species. Typical conditions would be 1 mg/mL TPA, 3 mM CMP sialic acid, 0.02 U/mL ST3Gal3, 32.degree. C. for 24 hours. Microbial growth can be halted either by sterile filtration or the inclusion of 0.02% sodium azide. The TPA concentration is most preferably in the range 0.1 mg/mL up to the solubility limit of the peptide. The concentration of CMP-SA should be sufficient for there to be excess over the available sites, and might range from 50 .mu.M up to 50 mM, and the temperature from 2.degree. C. up to 40.degree. C. The time required for complete reaction will depend on the temperature, the relative amounts of enzyme to acceptor substrate, the donor substrate concentration, and the pH. Other sialyltransferases that may be capable of adding sialic acid in 2,3 linkage include ST3Gal4; microbial transferases could also be used.

PGPUB-DOCUMENT-NUMBER: 20040230042

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040230042 A1

TITLE: Expression of class 2 mannosidase and class III mannosidase in lower eukaryotic cells

PUBLICATION-DATE: November 18, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Hamilton, Stephen	Enfield	NH	US	

APPL-NO: 10/ 616082

DATE FILED: July 8, 2003

RELATED-US-APPL-DATA:

child 10616082 A1 20030708

parent continuation-in-part-of 10371877 20030220 US PENDING

child 10371877 20030220 US

parent continuation-in-part-of 09892591 20010627 US PENDING

child 10616082 A1 20030708

parent continuation-in-part-of PCT/US02/41510 20021224 US PENDING

non-provisional-of-provisional 60214358 20000628 US

non-provisional-of-provisional 60215638 20000630 US

non-provisional-of-provisional 60279997 20010330 US

non-provisional-of-provisional 60344169 20011227 US

US-CL-CURRENT: 530/395, 435/254.2 , 435/471 , 435/69.1 , 536/23.5

ABSTRACT:

A method for producing human-like glycoproteins by expressing a Class 2 .alpha.-mannosidase having a substrate specificity for Man.alpha.1,3 and Man.alpha.1,6 glycosidic linkages in a lower eukaryote is disclosed. Hydrolysis of these linkages on oligosaccharides produces substrates for further N-glycan processing in the secretory pathway.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of U.S. application Ser. No. 10/371,877, filed on Feb. 20, 2003, which is a continuation-in-part of U.S. application Ser. No. 09/892,591, filed Jun. 27, 2001, which claims the benefit under 35 U.S.C. .sctn.119(e) of U.S. Provisional Application No. 60/214,358, filed Jun. 28, 2000, U.S. Provisional Application No. 60/215,638,

filed Jun. 30, 2000, and U.S. Provisional Application No. 60/279,997, filed Mar. 30, 2001, each of which is incorporated herein by reference in its entirety.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (20):

[0019] A significant number of proteins isolated from humans or animals are post-translationally modified, with glycosylation being one of the most significant modifications. An estimated 70% of all therapeutic proteins are glycosylated and thus currently rely on a production system (i.e., host cell) that is able to glycosylate in a manner similar to humans. Several studies have shown that glycosylation plays an important role in determining the (1) immunogenicity, (2) pharmnacokinetic properties, (3) trafficking and (4) efficacy of therapeutic proteins. It is thus not surprising that substantial efforts by the pharmaceutical industry have been directed at developing processes to obtain glycoproteins that are as "humanoid" or "human-like" as possible. To date, most glycoproteins are made in a mammalian host system. This may involve the genetic engineering of such mammalian cells to enhance the degree of sialylation (i.e., terminal addition of sialic acid) of proteins expressed by the cells, which is known to improve pharmacokinetic properties of such proteins. Alternatively, one may improve the degree of sialylation by in vitro addition of such sugars using known glycosyltransferases and their respective nucleotide sugars (e.g., 2,3-sialyltransferase and CMP-sialic acid).

PGPUB-DOCUMENT-NUMBER: 20040209802

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040209802 A1

TITLE: Treatment of disturbances of iron distribution

PUBLICATION-DATE: October 21, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Lehmann, Paul	Worms	DE	US	
Roeddiger, Ralf	Gorxheimertal	DE	US	
Walter-Matsui, Ruth	Altenbuseck	DE	US	

APPL-NO: 10/ 706701

DATE FILED: November 12, 2003

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	DOC-ID	APPL-DATE
EP	02026342.2	2002EP-02026342.2	November 22, 2002

US-CL-CURRENT: 514/12

ABSTRACT:

The present invention relates to the use of erythropoietin for the treatment of disturbances of iron distribution in heart diseases.

---------- KWIC ---------

Detail Description Paragraph - DETX (11):

[0017] Further, erythropoietin may be a glycoprotein analog having from 1 to 6 additional sites for glycosylation. Therefore, the present invention also relates to the use as described before, wherein the erythropoietin protein has the sequence of human erythropoietin modified by the addition of from 1 to 6 glycosylation sites. Glycosylation of a protein, with one or more oligosaccharide groups, occurs at specific locations along a polypeptide backbone and greatly affects the physical properties of the protein such as protein stability, secretion, subcellular localization, and biological activity. Glycosylation is usually of two types. O-linked oligosaccharides are attached to serine or threonine residues and N-linked oligosaccharides are attached to asparagine residues. One type of oligosaccharide found on both N-linked and O-linked oligosaccharides is N-acetylneuraminic acid (sialic acid), which is a family of amino sugars containing 9 or more carbon atoms. Sialic acid is usually the terminal residue on both N-linked and O-linked oligosaccharides and, because it bears a negative charge, confers acidic properties to the glycoprotein. Human erythropoietin, having 165 amino acids, contains three N-linked and one O-linked oligosaccharide chains which comprise about 40% of the total molecular weight of the glycoprotein. N-linked glycosylation occurs at asparagine residues located at positions 24, 38, and 83 and O-linked glycosylation occurs at a serine residue located at position 126. The oligosaccharide chains are modified with terminal sialic acid residues. Enzymatic removal of all sialic acid residues from the glycosylated erythropoietin results in loss of in vivo activity but not in vitro activity

because sialylation of erythropoietin prevents its binding, and subsequent clearance, by hepatic binding protein.

PGPUB-DOCUMENT-NUMBER: 20040176309

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040176309 A1

TITLE: Siglec inhibitors

PUBLICATION-DATE: September 9, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Kelm, Sorge	Lilienthal		DE	
Brossmer, Reinhard	Neckargemund		DE	

APPL-NO: 10/ 481529

DATE FILED: February 13, 2004

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	DOC-ID	APPL-DATE
DE	101 29 332.1	2001DE-101 29 332.1	June 19, 2001
DE	102 16 310.3	2002DE-102 16 310.3	April 12, 2002

PCT-DATA:
APPL-NO: PCT/EP02/06277
DATE-FILED: Jun 7, 2002
PUB-NO:
PUB-DATE:
371-DATE:
102(E)-DATE:

US-CL-CURRENT: 514/23, 514/100 , 514/326 , 514/328 , 514/432 , 514/460 , 514/89 , 536/108 , 546/22 , 546/242 , 549/28 , 549/417

ABSTRACT:

The invention relates to Siglec inhibitors that have an increased affinity for the receptor molecule. The Siglec inhibitors provided by the invention are preferably selective of a given Siglec molecule. The invention further relates to a method for producing Siglec inhibitors and to a method for increasing the binding selectivity for a given Siglec molecule. The invention also relates to pharmaceutical compositions that contain the Siglec inhibitors and to medical indications for the Siglec inhibitors.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (2):

[0002] Siglecs (sialic acid binding Ig-like lectins) are Ig-type lectins which are characterised by an N-terminal V-set domain which mediates the sialic acid bond. A varying number of Ig domains of the C2 set follows the Ig domain. Originally, the lectin family was found based on independent studies of sialoadhesin (siglec-1 CD 169), a macrophage lectin-like adhesion molecule and CD22 (siglec-2), a B-cell restricted member of the Ig superfamily (IgSF), which plays an important role in the regulation of the B-cell activation. It was also found that both molecules mediate the cell-cell interactions in vitro by

the detection of sialylated glycoconjugates. The cloning of sialoadhesin indicated high sequence similarities to CD22 and led to the conclusion that two further IgSF proteins, having a relationship in this respect, the myelin-associated glycoprotein (MAG/siglec-4) and CD33 (siglec-3), the binding of which to sialic acid was not previously known, also represent members of the siglec family. Six other human siglec molecules (siglecs 5-10) have been identified and characterised. These previously unknown molecules exhibit a high degree of sequence similarity to CD33 in their extracellular and intracellular domains and are collectively termed "siglecs standing in relation to CD33" (1; summary article). Reference (7) describes the cloning and characterisation of siglec-11 which is expressed from human dendritic cells.

PGPUB-DOCUMENT-NUMBER: 20040176279

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040176279 A1

TITLE: Flint glycoforms

PUBLICATION-DATE: September 9, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Jenkins, Nigel	Lausanne	IN	GB	
Witcher, Derrick Ryan	Fishers	IN	US	
Wroblewski, Victor John	Carmel		US	

APPL-NO: 10/ 466786

DATE FILED: April 26, 2004

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60265749 20010201 US

PCT-DATA:
APPL-NO: PCT/US02/00510
DATE-FILED: Jan 18, 2002
PUB-NO:
PUB-DATE:
371-DATE:
102(E)-DATE:

US-CL-CURRENT: 514/8

ABSTRACT:

The present invention provides FLINT isoforms having an average sialic acid content of 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 3.5, 4.0 or greater than 4.0 per molecule of FLINT. Also provided are mixtures of said isoforms, and methods for making said isoforms.

---------- KWIC ---------

Detail Description Paragraph - DETX (12):

[0023] "FLINT isoform" refers to sialic acid variants of FLINT. Native FLINT can occur with and accommodate 0, 1, 2, 3, 4, 5, or 6 sialic acids per molecule of FLINT, based on one N-linked site at Asn 144 of SEQ ID NO:1, and one O-linked site at Thr 174 of SEQ ID NO:1. A second O-linked site, at Thr 216 of SEQ ID NO:1, is substantially less glycosylated than the site at Thr 174. Protease-resistant analog R218Q can occur with 0, 1, 2, 3, 4, 5, 6, 7, or 8 sialic acids per molecule of protein, based on one N-linked site at Asn 144 of SEQ ID NO:1, one O-linked site at Thr 174 and one O-linked site at Thr 216 of SEQ ID NO:1. O-linked glycosylation is substantially enhanced at position 216 of R218Q (SEQ ID NO:1) when compared with native FLINT. The degree of sialylation will depend on the host cell and growth conditions. Sialylation can be enhanced in vitro using an enzymatic process described later in this

disclosure.

Detail Description Paragraph - DETX (39):
[0050] In Vitro Enhancement of Sialylation of FLINT

Detail Description Paragraph - DETX (40):
[0051] Also contemplated by the present invention is a method for enhancing the sialylation of FLINT by enzymatic modification in vitro.

PGPUB-DOCUMENT-NUMBER: 20040171826

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040171826 A1

TITLE: Endomannosidases in the modification of glycoproteins in eukaryotes

PUBLICATION-DATE: September 2, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Hamilton, Stephen	Enfield	NH	US	

APPL-NO: 10/ 695243

DATE FILED: October 27, 2003

RELATED-US-APPL-DATA:

child 10695243 A1 20031027

parent continuation-in-part-of 10371877 20030220 US PENDING

US-CL-CURRENT: 536/23.5, 435/254.2 , 435/483 , 435/69.1 , 530/395

ABSTRACT:

The present invention generally relates to methods of modifying the glycosylation structures of recombinant proteins expressed in fungi or other lower eukaryotes, to more closely resemble the glycosylation of proteins from higher mammals, in particular humans. The present invention also relates to novel enzymes and, nucleic acids encoding them and, hosts engineered to express the enzymes, methods for producing modified glycoproteins in hosts and modified glycoproteins so produced.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of U.S. application Ser. No. 10/371,877, filed on Feb. 20, 2003.

---------- KWIC ---------

Summary of Invention Paragraph - BSTX (8):

[0007] When proteins are isolated from humans or animals, a significant number of them are post-translationally modified, with glycosylation being one of the most significant modifications. Several studies have shown that glycosylation plays an important role in determining the (1) immunogenicity, (2) pharmacokinetic properties, (3) trafficking, and (4) efficacy of therapeutic proteins. An estimated 70% of all therapeutic proteins are glycosylated and thus currently rely on a production system (i.e., host) that is able to glycosylate in a manner similar to humans. To date, most glycoproteins are made in a mammalian host system. It is thus not surprising that substantial efforts by the pharmaceutical industry have been directed at developing processes to obtain glycoproteins that are as "humanoid" as possible. This may involve the genetic engineering of such mammalian cells to

enhance the degree of sialylation (i.e., terminal addition of sialic acid) of proteins expressed by the cells, which is known to improve pharmacokinetic properties of such proteins. Alternatively, one may improve the degree of sialylation by in vitro addition of such sugars by using known glycosyltransferases and their respective nucleotide sugar substrates (e.g. 2,3 sialyltransferase and CMP-Sialic acid).

PGPUB-DOCUMENT-NUMBER: 20040168208

PGPUB-FILING-TYPE: republication-amended

DOCUMENT-IDENTIFIER: US 20040168208 A2

TITLE: PRODUCTION OF BUTYRYLCHOLINESTERASES IN TRANSGENIC ANIMALS

PUBLICATION-DATE: August 26, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Karatzas, Costas	Quebec,		CA	
Huang, Yue-Jin	Quebec,		CA	
Lazaris, Anthoula	Quebec,		CA	

APPL-NO: 10/ 326892

DATE FILED: December 20, 2002

RELATED-US-APPL-DATA:

non-provisional-of-provisional 60/344,295 20011221 US

US-CL-CURRENT: 800/7, 800/14 , 800/18

ABSTRACT:

The present invention provides methods for the large-scale production of recombinant butyrylcholinesterase in cell culture, and in the milk and/or urine of transgenic mammals. The recombinant butyrylcholinesterases of this invention can be used to treat and/or prevent organophosphate pesticide poisoning, nerve gas poisoning, cocaine intoxication, and succinylcholine-induced apnea.

---------- KWIC ---------

Detail Description Paragraph - DETX (91):

[0084] As a means of producing recombinant BChE with a glycosylation profile that more closely resembles that of the native enzyme, the present invention is directed to transgenic animals that express both a BChE enzyme and one or more glycosyltransferases in their mammary glands and/or urinary endothelium, as well as cultured mammalian cells that express both a BChE enzyme and one or more glycosyltransferases. The presence of the glycosyltransferases in the intracellular secretory pathway of cells that are also expressing a secreted form of BChE catalyzes the transfer of glycan moieties to said BChE enzymes. The invention also encompasses addition of one or more glycosyltransferases to an in vitro reaction for the transfer of glycan moieties to a recombinant BChE produced by the transgenic animals or transfected mammalian cell lines of the invention. For example, recombinant BChE may be sialylated using the in vitro reaction conditions described in Chitlaru, et al. Biochem. J. (1998) 336:647-658. Thus, the glycosyltransferase which catalyzes transfer of glycans to the BChE enzyme may be expressed by the same cell that expresses the BChE enzyme, or the glycosyltransferase may be obtained from an external source and added to the recombinant BChE.

PGPUB-DOCUMENT-NUMBER: 20040147431

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040147431 A1

TITLE: Erythropoietin composition

PUBLICATION-DATE: July 29, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Papadimitriou, Apollon	Bichl		DE	

APPL-NO: 10/ 780297

DATE FILED: February 17, 2004

RELATED-US-APPL-DATA:

child 10780297 A1 20040217

parent continuation-of 09853731 20010511 US PENDING

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	DOC-ID	APPL-DATE
EP	00110355.5	2000EP-00110355.5	May 15, 2000

US-CL-CURRENT: 514/8

ABSTRACT:

The present invention relates to a liquid pharmaceutical composition consisting essentially of an erythropoietin protein, a multiple charged inorganic anion in a pharmaceutically acceptable buffer suitable to keep the solution pH in the range from about 5.5 to about 7.0, and optionally one or more pharmaceutically acceptable excipients. This composition is especially useful for the prophylaxis and treatment of diseases related to erythropoiesis.

PRIORITY TO RELATED APPLICATIONS

[0001] This application is a Continuation of Ser. No. 09/853,731, filed May 11, 2001, which is now pending.

---------- KWIC ---------

Detail Description Paragraph - DETX (13):

[0054] Further, the erythropoietin product of this invention may be a glycoprotein of the above sequences modified by having from 1 to 6 additional sites for glycosylation. Glycosylation of a protein, with one or more oligosaccharide groups, occurs at specific locations along a polypeptide backbone and greatly affects the physical properties of the protein such as protein stability, secretion, subcellular localization, and biological activity. Glycosylation is usually of two types. O-linked oligosaccharides are attached to serine or threonine residues and N-linked oligosaccharides are attached to asparagine residues. One type of oligosaccharide found on both N-linked and O-linked oligosaccharides is N-acetylneuraminic acid (sialic

acid), which is a family of amino sugars containing 9 or more carbon atoms. Sialic acid is usually the terminal residue on both N-linked and O-linked oligosaccharides and, because it bears a negative charge, confers acidic properties to the glycoprotein. Human erythropoietin, having 165 amino acids, contains three N-linked and one O-linked oligosaccharide chains which comprise about 40% of the total molecular weight of the glycoprotein. N-linked glycosylation occurs at asparagine residues located at positions 24, 38, and 83 and O-linked glycosylation occurs at a serine residue located at position 126. The oligosaccharide chains are modified with terminal sialic acid residues. Enzymatic removal of all sialic acid residues from the glycosylated erythropoietin results in loss of in vivo activity but not in vitro activity because sialylation of erythropoietin prevents its binding, and subsequent clearance, by hepatic binding protein.

PGPUB-DOCUMENT-NUMBER: 20040142856

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040142856 A1

TITLE: Glycoconjugation methods and proteins/peptides produced by the methods

PUBLICATION-DATE: July 22, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
DeFrees, Shawn	North Wales	PA	US	
Zopf, David	Wayne	PA	US	
Bayer, Robert	San Diego	CA	US	
Bowe, Caryn	Doylestown	PA	US	
Hakes, David	Willow Grove	PA	US	
Chen, Xi	Lansdale	PA	US	

APPL-NO: 10/ 410913

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10410913 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10410913 A1 20030409

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10410913 A1 20030409

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

non-provisional-of-provisional 60334692 20011121 US

non-provisional-of-provisional 60328523 20011010 US

US-CL-CURRENT: 514/8, 435/68.1

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (1059):
[1389] In vitro sialylation was carried out using the G2 glycoform antibody (1 mg/mL) by contacting it with 0.1 U/mg ST3Gal3 or 0.1 U/mg ST6Gal1, 5 mM CMP-sialic acid, at 32.degree. C. for 24 hr in a buffer of 50 mM Tris pH 7.4, 150 mM NaCl, and 3 mM CMP-SA. An aliquot was removed for glycan analysis, and the remaining products were affinity purified as described above.

Detail Description Paragraph - DETX (1332):
[1662] Sialylation. TPA expressed in mammalian cells will often contain a majority of the glycans terminating in sialic acid, but to ensure complete sialylation, it would be beneficial to first perform an in vitro sialylation. TPA in a suitable buffer (most preferably between pH 5.5 and 9, for example Tris buffered saline, pH 7.2) is incubated with CMP sialic acid and sialyltransferase for a time sufficient to convert any glycans lacking sialic acid to sialylated species. Typical conditions would be 1 mg/mL TPA, 3 mM CMP sialic acid, 0.02 U/mL ST3Gal3, 32.degree. C. for 24 hours. Microbial growth can be halted either by sterile filtration or the inclusion of 0.02% sodium azide. The TPA concentration is most preferably in the range 0.1 mg/mL up to the solubility limit of the peptide. The concentration of CMP-SA should be sufficient for there to be excess over the available sites, and might range from 50 .mu.M up to 50 mM, and the temperature from 2.degree. C. up to 40.degree. C. The time required for complete reaction will depend on the temperature, the relative amounts of enzyme to acceptor substrate, the donor substrate concentration, and the pH. Other sialyltransferases that may be capable of adding sialic acid in 2,3 linkage include ST3Gal4; microbial transferases could also be used.

PGPUB-DOCUMENT-NUMBER: 20040137557

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040137557 A1

TITLE: Remodeling and glycoconjugation of peptides

PUBLICATION-DATE: July 15, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
DeFrees, Shawn	North Wales	PA	US	
Zopf, David	Wayne	PA	US	
Bayer, Robert	San Diego	CA	US	
Bowe, Caryn	Doylestown	PA	US	
Hakes, David	Willow Grove	PA	US	
Chen, Xi	Lansdale	PA	US	

APPL-NO: 10/ 287994

DATE FILED: November 5, 2002

RELATED-US-APPL-DATA:

child 10287994 A1 20021105

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

US-CL-CURRENT: 435/68.1, 530/322

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group a peptide.

---------- KWIC ---------

Detail Description Paragraph - DETX (949):
[1803] Sialylation. TPA expressed in mammalian cells will often contain a majority of the glycans terminating in sialic acid, but to ensure complete sialylation, it would be beneficial to first perform an in vitro sialylation.

TPA in a suitable buffer (most preferably between pH 5.5 and 9, for example Tris buffered saline, pH 7.2) is incubated with CMP sialic acid and sialyltransferase for a time sufficient to convert any glycans lacking sialic acid to sialylated species. Typical conditions would be 1 mg/mL TPA, 3 mM CMP sialic acid, 0.02 U/mL ST3Gal3, 32.degree. C. for 24 hours. Microbial growth can be halted either by sterile filtration or the inclusion of 0.02% sodium azide. The TPA concentration is most preferably in the range 0.1 mg/mL up to the solubility limit of the peptide. The concentration of CMP-SA should be sufficient for there to be excess over the available sites, and might range from 50 .mu.M up to 50 mM, and the temperature from 2.degree. C. up to 40.degree. C. The time required for complete reaction will depend on the temperature, the relative amounts of enzyme to acceptor substrate, the donor substrate concentration, and the pH. Other sialyltransferases that may be capable of adding sialic acid in 2,3 .mu.linkage include ST3Gal4; microbial transferases could also be used.

US-PAT-NO: 6855544

DOCUMENT-IDENTIFIER: US 6855544 B1

TITLE: Recombinant protein production in a human cell

DATE-ISSUED: February 15, 2005

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Hateboer; Guus	Heemstede	N/A	N/A	NL
Verhulst; Karina Cornelia	Leiden	N/A	N/A	NL
Schouten; Govert Johan	Leiderdorp	N/A	N/A	NL
Uytdehaag; Alphonsus Gerardus	DeBilt	N/A	N/A	NL
Bout; Abraham	Moerkapelle	N/A	N/A	NL

APPL-NO: 09/ 549463

DATE FILED: April 14, 2000

PARENT-CASE:

RELATED APPLICATIONS

Under the provisions of 35 U.S.C. .sctn. 119(e), priority is claimed from Provisional Patent Application Serial No. 06/129,452 filed Apr. 15, 1999.

US-CL-CURRENT: 435/325, 435/320.1 , 435/455 , 435/69.1 , 435/69.7 , 536/23.1 , 536/23.5 , 536/23.72 , 536/24.1

ABSTRACT:

Methods and compositions for the production of recombinant proteins in a human cell line. The methods and positions are particularly useful for generating stable expression of human recombinant proteins of interest that are modified post-translationally, for example, by glycosylation. Such proteins may have advantageous properties in comparison with their counterparts produced in non-human systems such as Chinese Hamster Ovary cells.

28 Claims, 27 Drawing figures

Exemplary Claim Number: 1

Number of Drawing Sheets: 27

---------- KWIC ---------

Detailed Description Text - DETX (162):

The function of recombinant EPO in vivo is determined by its half-life in the bloodstream. Removal of EPO takes place by liver enzymes that bind to galactose residues in the glycans that are not protected by sialic acids and by removal through the kidney. Whether this filtering by the kidney is due to misfolding or due to under- or mis-glycosylation is unknown. Furthermore, EPO molecules that reach their targets in the bone marrow and bind to the EPO receptor on progenitor cells are also removed from circulation. Binding to the EPO receptor and down stream signalling depends heavily on a proper

glycosylation status of the EPO molecule. Sialic acids can, to some extent, inhibit binding of EPO to the EPO receptor, resulting in a lower effectivity of the protein. However, since the sialic acids prevent EPO from removal, these sugars are essential for its function to protect the protein on its travel to the EPO receptor. When sialic acids are removed from EPO in vitro, a better binding to the receptor occurs, resulting in a stronger down stream signalling. This means that the functionalities in vivo and in vitro are significantly different, although a proper EPO receptor binding property can be checked in vitro despite the possibility of an under-sialylation causing a short half-life in vivo (Takeuchi et al. 1989).

Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
L2	43	vitro near4 sialyla$	US-PGPUB; USPAT	OR	OFF	2005/04/04 14:38
L3	44	(commercial or scale or batch) near4 sialyl$	US-PGPUB; USPAT	OR	OFF	2005/04/04 15:00

PGPUB-DOCUMENT-NUMBER: 20050064540

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20050064540 A1

TITLE: Glycoprotein remodeling using endoglycanases

PUBLICATION-DATE: March 24, 2005

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Defrees, Shawn Ph.D	North Wales	PA	US	
Johnson, Karl F	Willow Grove	PA	US	

APPL-NO: 10/ 497284

DATE FILED: May 28, 2004

PCT-DATA:
APPL-NO: PCT/US02/38442
DATE-FILED: Nov 27, 2002
PUB-NO:
PUB-DATE:
371-DATE:
102(E)-DATE:

US-CL-CURRENT: 435/68.1, 530/395

ABSTRACT:

This invention provides methods for modifying glycosylation patterns of glycoproteins, including recombinantly produced glycoproteins. Also provided are glycoprotein compositions in which the glycoproteins have a homogeneous glycosylation pattern.

---------- KWIC ---------

Detail Description Paragraph - DETX (120):
[0163] Other sialyltransferases, including those listed in Table 4, are also useful in an economic and efficient large-scale process for sialylation of commercially important glycopeptides. As a simple test to find out the utility of these other enzymes, various amounts of each enzyme (1-100 mU/mg protein) are reacted with asialo-.alpha..sub.1 AGP (at 1-10 mg/ml) to compare the ability of the sialyltransferase of interest to sialylate glycopeptides relative to either bovine ST6Gal I, ST3Gal III or both sialyltransferases. Alternatively, other glycopeptides or glycopeptides, or N-linked oligosaccharides enzymatically released from the peptide backbone can be used in place of asialo-.alpha..sub.1 AGP for this evaluation. Sialyltransferases with the ability to sialylate N-linked oligosaccharides of glycopeptides more efficiently than ST6Gal I are useful in a practical large-scale process for peptide sialylation (as illustrated for ST3Gal III in this disclosure).

PGPUB-DOCUMENT-NUMBER: 20050032742

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20050032742 A1

TITLE: Chemo-enzymatic synthesis of sialylated oligosaccharides

PUBLICATION-DATE: February 10, 2005

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
DeFrees, Shawn	North Wales	PA	US	
McGuire, Edward J	Furlong	PA	US	

APPL-NO: 10/ 485892

DATE FILED: October 1, 2004

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	DOC-ID	APPL-DATE
US	60313278	2001US-60313278	August 17, 2001
US	60351444	2002US-60351444	January 23, 2002

PCT-DATA:
APPL-NO: PCT/US02/24574
DATE-FILED: Aug 1, 2002
PUB-NO:
PUB-DATE:
371-DATE:
102(E)-DATE:

US-CL-CURRENT: 514/54, 435/84 , 536/53

ABSTRACT:

In vitro/cell-free process of preparing a sialylated oligosaccharides are described. The sialylated oligosaccharides include gangliosides. The oligosaccharides linked to various moieties including sphingoids and ceramides. Novel compounds that comprise sphingoid groups are disclosed. The compounds include sialylated oligosaccharides including gangliosides as well as various sphingoids and ceramides.

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Detail Description Paragraph - DETX (170):
[0201] In some embodiments, the **sialylation methods used in the invention have increased commercial practicality through the use of bacterial sialyltransferases,** either recombinantly produced or produced in the native bacterial cells. Two bacterial sialyltransferases have been recently reported; an ST6Gal II from Photobacterium damsela (Yamamoto et al. (1996) J. Biochem. 120: 104-110) and an ST3Gal V from Neisseria meningitidis (Gilbert et al. (1996) J. Biol. Chem. 271: 28271-28276). The two recently described bacterial enzymes transfer sialic acid to the Gal.beta.1,4GlcNAc sequence on oligosaccharide substrates.

PGPUB-DOCUMENT-NUMBER: 20050031584

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20050031584 A1

TITLE: Interleukin-2:remodeling and glycoconjugation of interleukin-2

PUBLICATION-DATE: February 10, 2005

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
DeFrees, Shawn	North Wales	PA	US	
Zopf, David	Wayne	PA	US	
Bayer, Robert	San Diego	CA	US	
Bowe, Caryn	Doylestown	PA	US	
Hakes, David	Willow Grove	PA	US	
Chen, Xi	Lansdale	PA	US	

APPL-NO: 10/ 410980

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10410980 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10410980 A1 20030409

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10410980 A1 20030409

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

non-provisional-of-provisional 60344692 20011019 US

non-provisional-of-provisional 60328523 20011010 US

US-CL-CURRENT: 424/85.2, 530/351

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

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Detail Description Paragraph - DETX (667):
[0990] Other sialyltransferases, including those listed in Table 8, are also useful in an economic and efficient large-scale process for sialylation of commercially important glycopeptides. As a simple test to find out the utility of these other enzymes, various amounts of each enzyme (1-100 mU/mg protein) are reacted with asialo-.alpha..sub.1 AGP (at 1-10 mg/ml) to compare the ability of the sialyltransferase of interest to sialylate glycopeptides relative to either bovine ST6Gal I, ST3Gal III or both sialyltransferases. Alternatively, other glycopeptides or glycopeptides, or N-linked oligosaccharides enzymatically released from the peptide backbone can be used in place of asialo-.alpha..sub.1 AGP for this evaluation. Sialyltransferases with the ability to sialylate N-linked oligosaccharides of glycopeptides more efficiently than ST6Gal I are useful in a practical large-scale process for peptide sialylation (as illustrated for ST3Gal III in this disclosure).

Detail Description Paragraph - DETX (1046):
[1368] This Example demonstrates that sialylation of recombinant glycoproteins using the ST3 Gal III sialyltransferase required much less enzyme than expected. For a one kilogram scale reaction, approximately 7,000 units of the ST3Gal III sialyltransferase would be needed, instead of 100,000-150,000 units that earlier studies indicated. Purification of these enzymes from natural sources is prohibitive, with yields of only 1-10 units for a large scale preparation after 1-2 months work. Assuming that both the ST6Gal I and ST3Gal III sialyltransferases are produced as recombinant sialyltransferases, with equal levels of expression of the two enzymes being achieved, a fermentation scale 14-21 times greater (or more) would be required for the ST6Gal I sialyltransferase relative to the ST3Gal III sialyltransferase. For the ST6Gal I sialyltransferase, expression levels of 0.3 U/1 in yeast has been

reported (Borsig et al. (1995) Biochem. Biophys. Res. Commun. 210: 14-20). Expression levels of 1000 U/liter of the ST3 Gal III sialyltransferase have been achieved in Aspergillus niger. At current levels of expression 300-450,000 liters of yeast fermentation would be required to produce sufficient enzyme for sialylation of 1 kg of glycoprotein using the ST6Gal I sialyltransferase. In contrast, less than 10 liter fermentation of Aspergillus niger would be required for sialylation of 1 kg of glycoprotein using the ST3Gal III sialyltransferase. Thus, the fermentation capacity required to produce the ST3Gal III sialyltransferase for a large scale sialylation reaction would be 10-100 fold less than that required for producing the ST6Gal I; the cost of producing the sialyltransferase would be reduced proportionately.

PGPUB-DOCUMENT-NUMBER: 20040185146

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040185146 A1

TITLE: Methods for producing sialyloligosaccharides in a dairy source

PUBLICATION-DATE: September 23, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Pelletier, Marc	Doylestown	PA	US	
Barker, William A.	West Chester	PA	US	
Hakes, David J.	Willow Grove	PA	US	
Zopf, David A.	Strafford	PA	US	

APPL-NO: 10/ 798625

DATE FILED: March 11, 2004

RELATED-US-APPL-DATA:

child 10798625 A1 20040311

parent division-of 09955909 20010918 US GRANTED

parent-patent 6706497 US

child 09955909 20010918 US

parent continuation-of 08911393 19970814 US GRANTED

parent-patent 6323008 US

US-CL-CURRENT: 426/42

ABSTRACT:

The present invention provides methods for producing sialyloligosaccharides in situ in dairy sources and cheese processing waste streams, prior to, during, or after processing of the dairy source during the cheese manufacturing process. The methods of the present invention use the catalytic activity of .alpha.(2-3) trans-sialidases to exploit the high concentrations of lactose and .alpha.(2-3) sialosides which naturally occur in dairy sources and cheese processing waste streams to drive the enzymatic synthesis of .alpha.(2-3) sialyllactose. .alpha.(2-3) sialyloligosaccharides produced according to these methods are additionally encompassed by the present invention. The invention also provides for recovery of the sialyloligosaccharides produced by these methods. The invention further provides a method for producing .alpha.(2-3) sialyllactose. The invention additionally provides a method of enriching for .alpha.(2-3) sialyllactose in milk using transgenic mammals that express an .alpha.(2-3) trans-sialidase transgene. The invention also provides for recovery of the sialyllactose contained in the milk produced by this transgenic mammal either before or after processing of the milk. Transgenic mammals containing an .alpha.(2-3) trans-sialidase encoding sequence operably linked to a regulatory sequence of a gene expressed in mammary tissue are also provided by the

invention.

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Summary of Invention Paragraph - BSTX (26):
[0025] Numerous foreign proteins have successfully been transgenically expressed in the milk of livestock. Most of this work has focused on the expression of proteins which are foreign to the mammary gland. Colman, A., 1996, Am. J. Clin. Nutr. 63:639S-645S. To date, milk specific expression of transgenic livestock has been achieved through operably linking regulatory sequences of milk-specific protein genes to the target protein-encoding gene sequence, microinjecting these genetic constructs into the pronuclei of fertilized embryos, and implanting the embryos into recipient females. See e.g. Wright et al., 1991, Biotechnology (NY) 9:830-834; Carver et al., 1993, Biotechnology (NY) 11:1263-1270; Paterson et al., 1994, Appl. Microbiol. Biotechnol. 40:691-698. Proteins that have been successfully expressed in the milk of transgenic animals, include: .alpha.1-antitrypsin (Wright et al., 1991, Biotechnology (NY) 9:830-834; Carver et al., 1993, Biotechnology (NY) 11:1263-1270); Factor IX (Clark et al., 1989, Biotechnology (NY) 7:487-492); protein C (Velander et al., 1992, Proc. Natl. Acad. Sci. USA, 89:12003-12007); tissue plasminogen activator (Ebert et al., 1991, Biotechology (NY) 9:835-838); and fibrinogen. While most of these transgenes express proteins that supplement the composition of milk, very few, if any of the expressed proteins interact directly with the components of milk to alter the natural milk composition. There is a need for methods providing for the large scale production of .alpha.(2-3) sialyloligosaccharides, such as .alpha.(2-3) sialyllactose, which have commercial and/or therapeutic valve.

Summary of Invention Paragraph - BSTX (28):
[0026] The present invention greatly advances the field of commercial production of sialyloligosaccharides by providing methods for producing sialyloligosaccharides in situ in dairy sources and cheese processing waste streams. The methods of the invention have particular applications in producing .alpha.(2-3) sialyllactose in a dairy source-prior to, during, or after processing of the dairy source during the cheese manufacturing process, thereby greatly increasing the recoverable yield of .alpha.(2-3) sialyllactose from the dairy source.

PGPUB-DOCUMENT-NUMBER: 20040142856

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040142856 A1

TITLE: Glycoconjugation methods and proteins/peptides produced by the methods

PUBLICATION-DATE: July 22, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
DeFrees, Shawn	North Wales	PA	US	
Zopf, David	Wayne	PA	US	
Bayer, Robert	San Diego	CA	US	
Bowe, Caryn	Doylestown	PA	US	
Hakes, David	Willow Grove	PA	US	
Chen, Xi	Lansdale	PA	US	

APPL-NO: 10/ 410913

DATE FILED: April 9, 2003

RELATED-US-APPL-DATA:

child 10410913 A1 20030409

parent continuation-in-part-of 10360779 20030219 US PENDING

child 10410913 A1 20030409

parent continuation-in-part-of 10360770 20030106 US PENDING

child 10410913 A1 20030409

parent continuation-in-part-of 10287994 20021105 US PENDING

child 10287994 20021105 US

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

non-provisional-of-provisional 60334692 20011121 US

non-provisional-of-provisional 60328523 20011010 US

US-CL-CURRENT: 514/8, 435/68.1

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group to a peptide.

CROSS REFERENCE TO RELATED APPLICATIONS

[0001] This application is a continuation-in-part of prior Application No. PCT/US02/32263, filed Oct. 9, 2002; Provisional Patent Application No. 60/448,381, filed Feb. 19, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/438,582, filed Jan. 6, 2003 (converted to non-provisional application, same filing date, serial number not yet assigned); Provisional Patent Application No. 60/407,527, filed Aug. 28, 2002; Provisional Patent Application No. 60/404,249, filed Aug. 16, 2002; Provisional Patent Application No. 60/396,594, filed Jul. 17, 2002; Provisional Patent Application No. 60/391,777, filed Jun. 25, 2002; Provisional Patent Application No. 60/387,292, filed Jun. 7, 2002; Provisional Patent Application No. 60/334,301, filed Nov. 28, 2001; Provisional Patent Application No. 60/334,233, filed Nov. 28, 2001; Provisional Patent Application No. 60/344,692, filed Oct. 19, 2001; and Provisional Patent Application No. 60/328,523, filed Oct. 10, 2001.

---------- KWIC ---------

Detail Description Paragraph - DETX (667):
[0998] Other sialyltransferases, including those listed in Table 8, are also useful in an economic and efficient large-scale process for sialylation of commercially important glycopeptides. As a simple test to find out the utility of these other enzymes, various amounts of each enzyme (1-100 mU/mg protein) are reacted with asialo-.alpha..sub.1 AGP (at 1-10 mg/ml) to compare the ability of the sialyltransferase of interest to sialylate glycopeptides relative to either bovine ST6Gal I, ST3Gal III or both sialyltransferases. Alternatively, other glycopeptides or glycopeptides, or N-linked oligosaccharides enzymatically released from the peptide backbone can be used in place of asialo-.alpha..sub.1 AGP for this evaluation. Sialyltransferases with the ability to sialylate N-linked oligosaccharides of glycopeptides more efficiently than ST6Gal I are useful in a practical large-scale process for peptide sialylation (as illustrated for ST3Gal III in this disclosure).

Detail Description Paragraph - DETX (1046):
[1376] This Example demonstrates that sialylation of recombinant glycoproteins using the ST3 Gal III sialyltransferase required much less enzyme than expected. For a one kilogram scale reaction, approximately 7,000 units of the ST3Gal III sialyltransferase would be needed, instead of 100,000-150,000 units that earlier studies indicated. Purification of these enzymes from natural sources is prohibitive, with yields of only 1-10 units for a large scale preparation after 1-2 months work. Assuming that both the ST6Gal I and ST3Gal III sialyltransferases are produced as recombinant sialyltransferases, with equal levels of expression of the two enzymes being achieved, a fermentation scale 14-21 times greater (or more) would be required for the

ST6Gal I sialyltransferase relative to the ST3Gal III sialyltransferase. For the ST6Gal I sialyltransferase, expression levels of 0.3 U/l in yeast has been reported (Borsig et al. (1995) Biochem. Biophys. Res. Commun. 210: 14-20). Expression levels of 1000 U/liter of the ST3 Gal II sialyltransferase have been achieved in Aspergillus niger. At current levels of expression 300-450,000 liters of yeast fermentation would be required to produce sufficient enzyme for sialylation of 1 kg of glycoprotein using the ST6Gal I sialyltransferase. In contrast, less than 10 liter fermentation of Aspergillus niger would be required for sialylation of 1 kg of glycoprotein using the ST3Gal III sialyltransferase. Thus, the fermentation capacity required to produce the ST3Gal III sialyltransferase for a large scale sialylation reaction would be 10-100 fold less than that required for producing the ST6Gal I; the cost of producing the sialyltransferase would be reduced proportionately.

PGPUB-DOCUMENT-NUMBER: 20040137557

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040137557 A1

TITLE: Remodeling and glycoconjugation of peptides

PUBLICATION-DATE: July 15, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
DeFrees, Shawn	North Wales	PA	US	
Zopf, David	Wayne	PA	US	
Bayer, Robert	San Diego	CA	US	
Bowe, Caryn	Doylestown	PA	US	
Hakes, David	Willow Grove	PA	US	
Chen, Xi	Lansdale	PA	US	

APPL-NO: 10/ 287994

DATE FILED: November 5, 2002

RELATED-US-APPL-DATA:

child 10287994 A1 20021105

parent continuation-of PCT/US02/32263 20021009 US PENDING

non-provisional-of-provisional 60407527 20020828 US

non-provisional-of-provisional 60404249 20020816 US

non-provisional-of-provisional 60396594 20020717 US

non-provisional-of-provisional 60391777 20020625 US

non-provisional-of-provisional 60387292 20020607 US

non-provisional-of-provisional 60334301 20011128 US

non-provisional-of-provisional 60334233 20011128 US

US-CL-CURRENT: 435/68.1, 530/322

ABSTRACT:

The invention includes methods and compositions for remodeling a peptide molecule, including the addition or deletion of one or more glycosyl groups to a peptide, and/or the addition of a modifying group a peptide.

---------- KWIC ---------

Detail Description Paragraph - DETX (649):

[1504] Other sialyltransferases, including those listed in Table 7, are also useful in an economic and efficient large-scale process for sialylation of commercially important glycopeptides. As a simple test to find out the utility

of these other enzymes, various amounts of each enzyme (1-100 mU/mg protein) are reacted with asialo-.alpha..sub.1 AGP (at 1-10 mg/ml) to compare the ability of the sialyltransferase of interest to sialylate glycopeptides relative to either bovine ST6Gal I, ST3Gal III or both sialyltransferases. Alternatively, other glycopeptides or glycopeptides, or N-linked oligosaccharides enzymatically released from the peptide backbone can be used in place of asialo-.alpha..sub.1 AGP for this evaluation. Sialyltransferases with the ability to sialylate N-linked oligosaccharides of glycopeptides more efficiently than ST6Gal I are useful in a practical large-scale process for peptide sialylation (as illustrated for ST3Gal III in this disclosure).

Detail Description Paragraph - DETX (946):

[1800] This Example demonstrates that sialylation of recombinant glycoproteins using the ST3 Gal III sialyltransferase required much less enzyme than expected. For a one kilogram scale reaction, approximately 7,000 units of the ST3Gal III sialyltransferase would be needed, instead of 100,000-150,000 units that earlier studies indicated. Purification of these enzymes from natural sources is prohibitive, with yields of only 1-10 units for a large scale preparation after 1-2 months work. Assuming that both the ST6Gal I and ST3Gal III sialyltransferases are produced as recombinant sialyltransferases, with equal levels of expression of the two enzymes being achieved, a fermentation scale 14-21 times greater (or more) would be required for the ST6Gal I sialyltransferase relative to the ST3Gal III sialyltransferase. For the ST6Gal I sialyltransferase, expression levels of 0.3 U/1 in yeast has been reported (Borsig et al. (1995) Biochem. Biophys. Res. Commun. 210: 14-20). Expression levels of 1000 U/liter of the ST3 Gal III sialyltransferase have been achieved in Aspergillus niger. At current levels of expression 300-450,000 liters of yeast fermentation would be required to produce sufficient enzyme for sialylation of 1 kg of glycoprotein using the ST6Gal I sialyltransferase. In contrast, less than 10 liter fermentation of Aspergillus niger would be required for sialylation of 1 kg of glycoprotein using the ST3Gal III sialyltransferase. Thus, the fermentation capacity required to produce the ST3Gal III sialyltransferase for a large scale sialylation reaction would be 10-100 fold less than that required for producing the ST6Gal I; the cost of producing the sialyltransferase would be reduced proportionately.